ABSTRACT OF THE DISCLOSURE

The invention provides a method of conferring
donor CDR binding affinity onto an antibody acceptor
variable region framework. The method consists of:
5   (a) constructing a population of altered antibody
variable region encoding nucleic acids, said population
comprising encoding nucleic acids for an acceptor
variable region framework containing a plurality of
different amino acids at one or more acceptor framework
10   region amino acid positions and donor CDRs containing a
plurality of different amino acids at one or more donor
CDR amino acid positions; (b) expressing said population
of altered variable region encoding nucleic acids, and
(c) identifying one or more altered variable regions
15   having binding affinity substantially the same or greater
than the donor CDR variable region. The acceptor
variable region framework can be a heavy or light chain
variable region framework and the populations of heavy
and light chain altered variable regions can be expressed
20   alone to identify heavy or light chains having binding
affinity substantially the same or greater than the donor
CDR variable region. The populations of heavy and light
chains additionally can be coexpressed to identify
heteromeric altered variable region binding fragments.
25   The invention also provides a method of simultaneously
grafting and optimizing the binding affinity of a
variable region binding fragment. The method consists
of: (a) constructing a population of altered heavy chain
variable region encoding nucleic acids comprising an
30   acceptor variable region framework containing donor CDRs
and a plurality of different amino acids at one or more
framework region and CDR amino acid positions;
(b) constructing a population of altered light chain

variable region encoding nucleic acids comprising an acceptor variable region framework containing donor CDRs and a plurality of different amino acids at one or more framework regions and CDR amino acid positions;

5   (c) coexpressing said populations of heavy and light chain variable region encoding nucleic acids to produce diverse combinations of heteromeric variable region binding fragments, and (d) identifying one or more heteromeric variable region binding fragments having

10 affinity substantially the same or greater than the donor CDR heteromeric variable region binding fragment. A method of optimizing the binding affinity of an antibody variable region is also provided. The method consists of: (a) constructing a population of antibody variable

15 region encoding nucleic acids, said population comprising two or more CDRs containing a plurality of different amino acids at one or more CDR amino acid positions; (b) expressing said population of variable region encoding nucleic acids, and (c) identifying one or more

20 variable regions having binding affinity substantially the same or greater than the donor CDR variable region. The variable region populations can be heavy or light chains and can be expressed as individual populations or they can be coexpressed to produce heteromeric variable

25 region binding fragments.